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THE UNIVERSITY OF ALBERTA

FATE OF LABELLED NITROGEN FERTILIZER IN LIMED, ELEMENTAL SULPHUR - LADEN FOREST SOILS

BY

DAVID ALLAN GOWER

A THESIS

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IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

IN

SOIL- PLANT RELATIONSHIPS

DEPARTMENT OF SOIL SCIENCE

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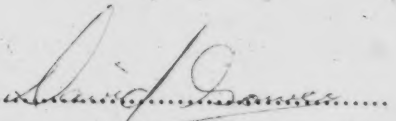

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FATE OF LABELLED NITROGEN FERTILIZER IN LIMED, ELEMENTAL SULPHUR - LADEN FOREST SOILS

submitted by

DAVID GOWER

in partial fulfilment of the requirements for the degree of

MASTER OF SCIENCE

in

SOIL-PLANT RELATIONSHIPS

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Supervisor

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Date: May 14, 1981

ABSTRACT

Nitrogen fertilization is an important step in reclaiming soil acidified by windblown elemental sulphur (S^o). The fate of ^{15}N fertilizer in previously limed, S^o-laden, forested Gray Luvisolic soils was investigated in growth chamber and field experiments. For the growth chamber study, $(^{15}NH_4)_2SO_4$ was added to nonplanted and planted potted soil containing three levels of S^o and lime: none (a control); medium; and high (HLS). During the 14 week incubation the HLS treatment had the highest: rate of S^o oxidation and N immobilization; flush of N from $CHCl_3$ fumigation; and accumulation of NO_3^-. The flush of N was correlated ($r^2 = 0.97$) with the disappearance of S^o in the HLS treatment. Compared to the control, HLS plants were more N deficient, having a lower: percentage of N derived from fertilizer (28 versus 54%); shoot yield (3.1 versus 3.8g per pot); and shoot to root ratio (0.5 versus 1.0). In the field investigation, the fate of NH_4^+ fertilizer was compared: 1) with NO_3^- fertilizer, and 2) among treatments with four different levels of S^o and lime (control; HLS; no lime plus medium S^o (NLS); and air applied lime plus medium S^o (ALS). Solutions of $(^{15}NH_4)_2SO_4$ or $K^{15}NO_3$ (90 kg N ha^{-1}) were added to cylinders, and a grass seed mixture sown. Eighty days after fertilizer application, the NH_4^+ fertilized soils, relative to NO_3^- fertilized, had higher amounts of: total recovered ^{15}N (86 versus 40%); immobilized ^{15}N (55 versus 34%); and unlabelled mineral N (64 versus 28 kg ha^{-1}). In the other field trial, the more acid ALS and NLS, relative to HLS, had greater amounts of nonlabelled mineral N (72 versus 25 kg ha^{-1}), and ^{15}N below the 10 cm depth of mineral soil (35 versus 15%); and less immobilized ^{15}N (57 versus 66%). Plants only grew on the control and HLS treatments and the plant recovery of ^{15}N was only 2 to 4%. Nitrification occurred, even though the pH was 4 or less. This study suggests reclamation will be more effective if fertilizer N is applied: 1) in greater amounts to soils with extremely high amounts of S^o and lime; 2) as NH_4^+, instead of NO_3^-; and 3) using frequent light applications, rather than a single large dose.

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1. INTRODUCTION

Elemental sulphur (S^o) is a by-product of sour natural gas processing. During treatment of S-containing natural gas, H_2S is removed, converted to S^o, and stored in large outdoor blocks. For over a decade wind erosion and mechanical breakup of the blocks has led to deposition of S^o in the immediate surroundings. Oxidation of S^o to H_2SO_4 by sulphur oxidizing microorganisms in the soil has resulted in severe soil acidification and loss of understory vegetation (Gal, 1986; Kennedy et al., 1985; Maynard et al., 1986; Nyborg, 1982). This problem is not as serious as it once was because S^o handling procedures have improved and the amount of stored S^o is declining due to increased world demand. Some large gas processing plants are now equipped with towers that prill S^o. These may reduce the intensity of deposition, but increase the area exposed to windblown S^o.

In this study a forested Gray Luvisolic soil acidfied by S^o was investigated. The site was located in the foothills, approximately 50 km SW of Rocky Mountain House, Alberta (52°13'N, 115°10'W). Several characteristics of this S^o-affected area were examined by Addison et al. (1984), Kennedy et al. (1985), and Maynard et al. (1983, 1986). Soils near the S^o storage block were compared to nearby sites with little or no S^o deposition. Higher total, elemental, and extractable S contents; and lower pH, extractable cations, and CO_2 respiration were found in soil less than 100 m from the block. The effects were greatest in the LFH horizons. Near the S^o block total vegetative cover and diversity were reduced and bryophytes were no longer present .

Reclamation of soils acidified by S^o has entailed adding high rates of $CaCO_3$ (lime) and N fertilizer (Nyborg, 1982). In agricultural systems these measures resulted in plants with normal nutrient contents (Bertrand, 1973). In forest soil, however, attempts to

establish grasses prior to native species invasion have met with limited success (Gal, 1986; Nyborg, 1983). Apparently this was due to the rapid disappearance of N fertilizer from the mineral form, as little growth occurred unless repeated N applications were made (Nyborg, 1982). The specific reasons for this N deficiency have not been clarified.

The purpose of this study was to identify the primary processes responsible for reduction of N available to plants in S^o-laden soil. Nyborg (1982) has suggested that the autotrophic S^o oxidizers found in these soils (Germida et al., 1985; Maynard et al., 1986) may be responsible for the reduction in plant available N. With a steady energy source supplied by S^o, N would be needed for growth of the organisms, and significant amounts of N could be immobilized. This could deprive plants of N and reduce the chances for successful reclamation of the S^o-affected area. In the first paper of this thesis N dynamics were examined during incubation of N fertilized S^o-laden soil. Emphasis was placed on N immobilization and its influence on plant growth. The second paper consisted of two field experiments. Little work has been done on the most suitable type of N fertilizer to apply to S^o-laden soil. In one field trial the potential effectiveness of ammonium and nitrate-based N fertilizers was compared. The second field experiment determined the effect of four different levels of S^o and lime on the fate of NH_4^+ fertilizer. The results of this research allowed formulation of recommendations regarding the reclamation of S^o-laden soil.

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2. GROWTH CHAMBER EXPERIMENT

INTRODUCTION

Reclamation of soils acidified by S^o has entailed adding high rates of $CaCO_3$ (lime) and N fertilizer (Nyborg, 1982). In forest soil, attempts to establish grasses prior to native species invasion have met with limited success (Gal, 1986; Nyborg, 1983). Added fertilizer N disappeared rapidly from the mineral form and little growth occurred unless repeated N applications were made (Nyborg, 1982). Autotrophic S^o oxidizers, identified as Thiobacillus thiooxidans and T. thioparus (Maynard et al., 1986) may be partially responsible for the reduction in plant available N. With a steady energy source supplied by S^o, the organisms could immobilize significant amounts of N. Heterotrophs could also immobilize more N given the more favorable pH after liming. Another possible mechanism of mineral N removal in these soils is increased gaseous evolution (Gal, 1986).

The purpose of this study was to identify the relative importance of processes responsible for the reduction of plant available fertilizer N in limed, S^o-laden soil. In this incubation experiment, the fate of fertilizer N was compared in forest soils with different levels of S^o and lime. Two hypotheses were tested: 1) that net immobilization of added ^{15}N would be greater in soil with higher amounts of S^o and lime; and consequently, 2) plant yield and uptake of ^{15}N would be less due to the higher N immobilization.

MATERIALS AND METHODS

Field site and sampling

Soils were obtained from an area adjacent to a sour gas plant located approximately 50 km southwest of Rocky Mountain House, Alberta (52°13'N, 115°10'W). The site has an elevation of 1200m and is part of the Lower Foothills section of the Boreal Forest (Rowe, 1972). *Pinus contorta* Loudon var. *latifolia* Engelm. and *Populus tremuloides* Michx. trees dominate the canopy, and Brunisolic and Podzolic Gray Luvisols are the common soil types (Addison et al., 1984). Further information on the soil and vegetation of the area is available from Addison et al. (1984). The soils used in this study were assumed to have a similar history, except with respect to previous additions of S^o and lime.

Three treatments were sampled in 1985 from previously established plots: a control; a soil with a high content of lime and S^o (HLS), and one with medium levels of lime and S^o (MLS) (Table 2.1). Neither lime nor S^o were added to the soil at or after field sampling. The control site had received little or no S^o and no lime. The other 2 treatments had been exposed to significant S^o dusting for about 5 years and had received several applications of lime ($CaCO_3$) from 1980 to 1984. Additions applied by aircraft totalled 22,170 kg ha^{-1}; most of this was in the form of coarsely (71% > 0.24 mm) ground limestone. Hand applied lime additions totalled 7,750 kg ha^{-1} of finely (8% > 0.24 mm) ground limestone. The finer lime is more effective in increasing soil pH because it has greater surface contact with the soil. In addition, the HLS, but not the MLS treatment, received an extra hand broadcast application of finely ground $CaCO_3$ (12,080 kg ha^{-1}) and finely divided S^o (2,240 kg ha^{-1}) in the fall of 1980 (Gal, 1986).

Table 2.1. Treatment locations and soil properties.

Treatment	Distance to S^o Block	S^o Additions	Lime Applied	pH	S^o	C	N
	m		t ha^{-1}		mg kg^{-1}	----------%	----------
Control	1500	None	None	4.4	50	4.4	0.16
High Lime, High S^o	250	Windblown + 2.2 t ha^{-1}	22.1 coarse + 19.8 fine	6.3	6,200	5.4	0.17
Medium Lime and S^o	250	Windblown	22.1 coarse + 7.8 fine	5.5	2,300	4.5	0.15

For each treatment, soil from three replicates approximately 15 m apart was sampled and combined. For each replicate, understory vegetation and green moss were removed, and a 30 x 40 cm area was excavated to a depth of 10 cm into the mineral soil. The soil was passed through a 0.64 cm sieve, mixed, and stored at 4°C until use. The mixing of organic (average depth of 6 cm) and mineral layers of soil represented a potential reclamation technique which would facilitate the incorporation of lime into the soil.

Experimental design

The fate of fertilizer N in the three soils was followed during a 14-week incubation experiment in a growth chamber. One week prior to fertilizer addition, moist soil (equivalent to 700 g of oven dry soil) was added to each of 66 pots (22 pots per soil). The pots had a diameter of 14.5 cm at the top, a depth of 10 cm, and a volume of 1500 cm^3. There were 2 subtreatments for each soil: without plants, and with plants. Nonplanted pots were destructively sampled at 0, 2, 8, and 14 weeks. The time 0 sampling was conducted prior to fertilizer addition. The planted pots were sampled at 8 and 14 weeks. There were 4 replicates per treatment for each sampling time, except for the planted pots at 14 weeks, which had two replicates per treatment. Fertilizer solutions were mixed into the soil at the following rates: 154 mg kg^{-1} or 70 kg ha^{-1} of N as $(^{15}NH_4)_2SO_4$ (4.94% atom excess), 22 mg kg^{-1} or 10 kg ha^{-1} of P as NaH_2PO_4, and 44 mg kg^{-1} or 20 kg ha^{-1} of K as KCl. Twenty *Lolium perenne* L. seeds were sown in each planted pot, and after 10 days plants were thinned to 8 per pot. Pots were placed in a growth chamber under alternating 12 hour cycles of light (one third fluorescent plus full incandescent light, 20°C), and dark (15°C). Soil moisture contents were kept between 70 and 100% of field capacity throughout the experiment. The field capacity was determined by wetting the top potion of a column of soil, letting it drain freely, and measuring the moisture content after 24 hr (Shaw 1927).

Sampling and analytical procedures

Each pot of soil was processed as follows. Moist soil was passed through a 2 mm sieve. The equivalent of approximately 25 g of oven dry soil were fumigated for 24 hr with chloroform and immediately extracted (Brookes et al., 1985). Both nonfumigated and fumigated soil were extracted with 0.5M K_2SO_4 for 1 hr (1g soil:4mL solution ratio). For the planted pots mineral N values were low, consequently a 1:2 ratio was employed. Extracts were suction filtered through Whatman no. 42 filter paper and stored at -15°C. Kjeldahl digestion ($KMnO_4$ pretreatment to include NO_2^- and NO_3^-; Bremner and Mulvaney 1982) of the K_2SO_4 extract, followed by steam distillation, provided total soluble N (TSN). Three heating stages were used in the Kjeldahl digestion. The first was at low temperature and drove off most of the water, and the time of digestion depended on the amount of extract available. When approximately 20 mL of liquid were left, the temperature was elevated to 220°C for three hours, and finally to 360°C for 5 hours. The flush of N was determined as the difference between TSN in the nonfumigated and fumigated soil (Brookes et al., 1985). This is an index of microbial biomass N. For the nonfumigated extract, NH_4^+ and NO_3^- were determined separately by steam distillation (Bremner, 1965). Subtraction of mineral N from TSN yielded soluble organic N (SON). Nonextracted soil was air dried. For planted pots, shoots were clipped at the soil surface and stored at -15°C. Roots were later washed free of air dried soil over a 2 mm sieve, and, like shoots, dried at 70°C, weighed, and ground. Total N and ^{15}N abundances were measured by combustion of air dried finely ground soil and plant samples in a Sira 1500 Automatic N analyzer coupled with a V. G. Isogas continuous flow mass spectrometer. Residual organic N was calculated as the difference between total N and the sum of mineral N plus flush of N.

Distillates derived from NH_4^+ and NO_3^- were dried and saved for ^{15}N analysis. For analysis of the $SO^{15}N$ fraction, extracts from which NH_4^+ and NO_3^- had been distilled were filtered (no. 1 Whatman paper), digested, and redistilled. The ^{15}N excess of the flush was calculated by applying the isotope dilution equation (Hauck, 1982) to the TSN fractions before and after fumigation:

$$\% \text{ excess flush} = \frac{\% \text{ excess TSN after (mg TSN after)} - \% \text{ excess TSN before (mg TSN before)}}{\text{mg flush}}$$

Where the ratio of mg of TSN before, to mg of flush was greater than 4:1, this calculation was not performed, because the errors in determining the ^{15}N excess of a small pool from two larger ones become too great. Hauck (1982) recommends a maximum ratio of 5:1. For the residual organic N fraction, the recovery of labelled N was determined by the difference between total N, and the sum of mineral N plus flush of N.

The turbidimetric method of Hart (1961) was employed to determine the S^o content of soil. Soil pH was measured in 0.01M $CaCl_2$ (1g soil:2mL solution ratio). Total C was measured by dry combustion in a Leco furnace (Nelson and Sommers, 1982).

Statistical analysis

ANOVA was performed to analyze: 1) the effect of nonplanted treatments (2 way - 3 treatments X 3 times); 2) the effect of planted treatments (2 way - 3 treatments X 2 times); 3) planted vs nonplanted soil (3 way - 3 treatments X 2 subtreatments X 2 times); and 4) plant material (2 way - 3 treatments X 2 times). For ANOVA 3), variances were heterogeneous in some cases. These parameters were (with the corrective transformation in brackets): NH_4^+-N (square root); TSN (square root); and

the % of ^{15}N recovered in: NH_4^+-N (log), TSN (reciprocal), and flush (log). The LSD test was used to compare means for planned comparisons, ie. - among the 3 treatments within a sampling time.

RESULTS

S^0 and N dynamics in nonplanted soil

Initially the HLS treatment had the highest S^0 content (Fig. 2.1a) and pH (Fig. 2.1b), while the control had the lowest. There was a concomitant decline in S^0 content and pH in the HLS and MLS treatments. This decrease was more rapid in the HLS treatment. The control showed little change in these variables. Prior to fertilizer addition mineral N (NH_4^+-N plus NO_3^--N) was less than 2 mg kg^{-1} in the three treatments. While NH_4^+ (Fig. 2.1c) declined from 2 to 14 weeks in all cases, it was always lowest in the HLS treatment. Little NO_3^- accumulated during the experiment. In the control and the MLS treatment NO_3^--N remained below 2 mg kg^{-1}, and constituted less than 6% of the mineral N (data not shown). In the HLS treatment similarly low levels of NO_3^--N were found through 8 weeks, but after 14 weeks, NO_3^--N increased to 13 mg kg^{-1} or 38% of the mineral N (Fig. 2.1c). Initially the flush of N, which provides an index of microbial biomass N, increased in the HLS treatment, but declined in the control (Fig. 2.1d) . At 14 weeks there was no significant difference in flush size among the treatments. When planted and nonplanted data were combined the flush size was correlated with the decrease in S^0 (N flush = 22.0 + .00411 x S^0 oxidized; $r=0.98$, $p\leq0.01$, $n=5$) for the HLS treatment, but not for the MLS treatment.

Soluble organic N (SON) patterns were variable among the 3 treatments (Fig. 2.1e). When nonplanted and planted data was combined, SON was inversely related to flush for the control ($r=-0.52$, $p\leq0.05$, $n=18$) and MLS treatments ($r=-0.69$, $p\leq0.01$ $n=18$). In contrast, a positive correlation between % ^{15}N excess in the SON and flush fractions was found for all 3 treatments ($r=0.66$, $p\leq0.01$, $n=40$).

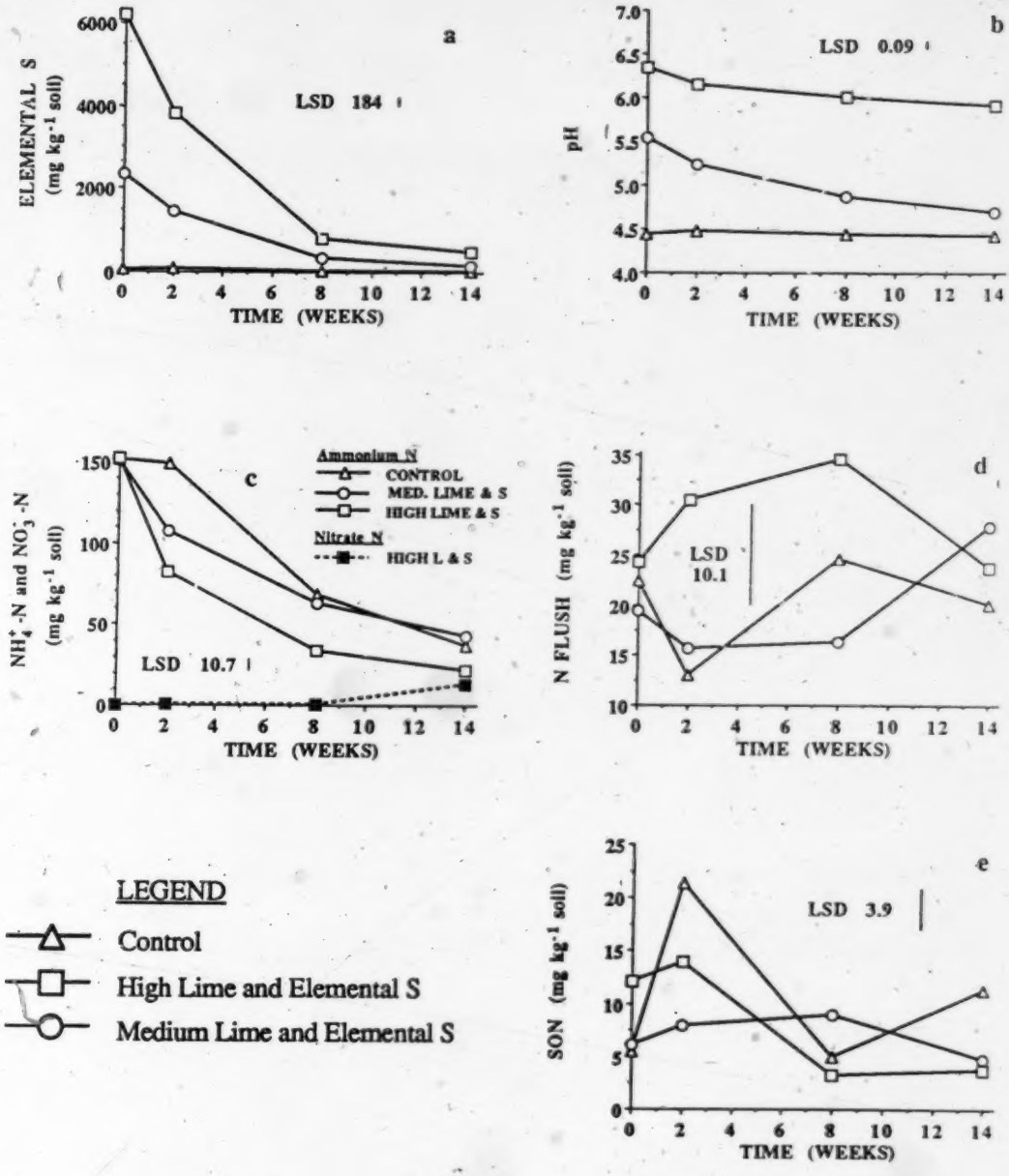

Figure 2.1. Changes in pH, S^o, and N in nonplanted soil at 2, 8, and 14 weeks.
a) Elemental S; b) pH; c) NH_4^+-N and NO_3^--N;
d) Flush of N from chloroform fumigation; e) Soluble organic N
Time zero NH_4^+-N includes added fertilizer N (154 mg kg^{-1})

Labelled N data confirmed the findings for nonlabelled N. Recovery of ^{15}N (Table 2.2) in the NH_4^+ fraction declined, and this corresponded to an increased recovery in residual organic N. This pattern was strongest in the HLS treatment, which had more rapid rates of ^{15}N disappearance in the NH_4^+ fraction, and buildup in the residual organic N. Generally, the HLS treatment had the highest recoveries in residual N, and lowest in mineral forms. The greatest recoveries in mineral forms at 8 and 14 weeks were found in the MLS treatment.

Unrecovered labelled N ranged from 9 to 16% (Table 2.2). However, repeated tests using freshly prepared ^{15}N-labelled fertilizer solution added to air dry soil of all three treatments resulted in 89% recovery of labelled N. The reason for this uniformly repeatable low recovery apparently resided with the mass spectrometer. When total recoveries were normalized to 89%, missing N ranged from 0 to 6%. In the HLS treatment unaccounted for ^{15}N averaged 4% greater ($p \leq 0.05$) than the control.

Plant growth and the influence of plants

Nonlabelled N, pH, and S^o for planted soil are shown in Table A.1. Results for ^{15}N in planted soil showed similarities to those of nonplanted soil (Table 2.2 versus 2.3). Again the HLS treatment had the highest recoveries in the residual and flush forms. Consequently, plant recovery of labelled N was lowest in this treatment. At 8 weeks, recovery in the residual fraction was greatest in the MLS treatment. Unadjusted unrecovered ^{15}N ranged from 9 to 16%, and was significantly higher (4% more) in the HLS treatment. Generally planted soils had less labelled and nonlabelled N in soluble forms than nonplanted, but total N recovery did not differ significantly (Table 2.4). Even though plant uptake kept mineral N at 5 mg kg^{-1} or less, the flush of N was not

Table 2.2. Percent of labelled N recovered in soil N fractions for nonplanted soil.

N form	Time (weeks) 2	8	14
	Control		
NH_4^+	77.4	20.2	7.3
NO_3^-	ND	0.4	0.1
Flush of N†	ND	6.2	3.5
Residual Organic N‡	11.6§	61.8	80.2
Total Recovery	89.0	88.6	91.1
	High Lime and S^o		
NH_4^+	33.7	7.8	4.1
NO_3^-	ND	0.1	2.1
Flush of N†	3.6	7.4	4.3
Residual Organic N‡	50.2	68.9	77.5
Total Recovery	86.5	84.2	88.0
	Medium Lime and S^o		
NH_4^+	58.3	23.4	12.6
NO_3^-	ND	0.3	0.2
Flush of N†	ND	4.9	4.8
Residual Organic N‡	30.7§	55.7	71.2
Total Recovery	89.0	84.3	88.8

Statistical analysis for nonplanted pots

	Source of variation			LSD¶
	Treatment	Time	Trt x Time	($p < 0.05$)
NH_4^+	**	**	**	5.7
NO_3^-	**	**	**	0.4
Flush of N	ns	*	ns	ns
Residual Organic N	**	**	**	7.6
Total Recovery	*	**	ns	ns

*, **, ns significant at $p < 0.05$, 0.01, and nonsignificant, respectively

ND not determined

† uncorrected for microbial biomass N

‡ total N - (flush of N + mineral N)

§ includes flush of N

¶ LSDs are for comparisons within a time only

Table 2.3. Percent of labelled N recovered in soil N fractions and plants for planted soil.†

N form	Time (weeks) 8	14
	Control	
NH_4^+-N	0.3	0.0
NO_3^--N	0.0	0.0
Flush of N	4.1	2.7
Residual Organic N	52.3	52.4
Plants	34.6	33.9
Total Recovery	91.3	89.0
	High Lime and S^o	
NH_4^+-N	0.2	0.0
NO_3^--N	0.0	0.0
Flush of N	6.1	3.9
Residual Organic N	63.8	58.7
Plants	16.2	21.3
Total Recovery	86.3	83.9
	Medium Lime and S^o	
NH_4^+-N	1.8	0.0
NO_3^--N	0.0	0.0
Flush of N	4.0	3.4
Residual Organic N	46.7	45.1
Plants	36.2	41.2
Total Recovery	88.7	89.7

Statistical analysis for planted pots

	Source of variation			LSD ($p < 0.05$)	
	Treatment	Time	Trt x Time	8 wks	14 wks
NH_4^+-N	ns	*	ns	ns	ns
NO_3^--N	ND	ND	ND	ND	ND
Flush of N	**	**	ns	ns	ns
Residual Organic N	**	*	ns	ns	ns
Plants	**	**	**	3.2	4.5
Total Recovery	*	ns	ns	ns	ns

† Footnotes are the same as for Table 2.2

Table 2.4. Comparison of N pools, S^o, and pH in planted versus nonplanted soil.†

	Nonlabelled			Labelled		
Parameter	Nonplanted	Planted	Significance	Nonplanted	Planted	Significance
Nitrogen	---mg N kg^{-1} soil---			-----% recovery-----		
NH_4^+-N	44.4	1.3	**	12.6	0.4	**
NO_3^--N	3.2	0.1	**	ND	ND	N/A
Soluble Organic	6.2	4.7	*	0.5	0.3	**
Total Soluble	53.7	6.1	**	13.0	0.6	**
Flush	24.5	25.4	ns	5.2	4.1	*
Residual Organic	1653	1640	ns	69.4	52.9	**
Total	1701	1641	**	87.9	88.8	ns
Elemental S (mg S kg^{-1} soil)	305	436	**			
pH	5.1	4.9	**			

*, **, ns significant at $p < 0.05$, 0.01, and nonsignificant, respectively (significant differences hold for both 8 and 14 weeks, except for SON)

N/A not applicable

ND not determined

† data averaged across treatments and times

lower than in nonplanted soil. Elemental S and pH were significantly different in planted compared to nonplanted soil.

Plants (Table 2.5) grown in the HLS treatment had the lowest: shoot yield, shoot to root ratio, and N derived from fertilizer. At 8 weeks the HLS treatment plants had the least % N in both roots and shoots, but there was no difference among treatments at 14 weeks. At 14 weeks root mass was greatest in the HLS treatment, and least in the control.

Table 2.5. N, yield, and N derived from fertilizer for plants at 8 and 14 weeks.

Portion of Plant	Parameter	Time (weeks)						Statistical Analysis		
		8	14	8	14	8	14	significance of treatment	LSD ($p < 0.05$) 8	LSD ($p < 0.05$) 14
		Control		High Lime and S^o		Medium Lime and S^o				
Shoots	N (%)	2.51	1.02	2.15	1.04	2.68	0.92	ns	0.30	0.43
	Mass (g)	1.92	3.79	1.37	3.06	1.96	4.60	**	0.26	0.37
	Ndff† (%)	59.6	58.3	44.6	42.7	64.2	61.6	**	ns	ns
Roots	N (%)	1.14	0.75	0.91	0.75	1.12	0.74	*	0.12	0.09
	Mass (g)	1.47	3.91	1.55	6.50	1.06	5.44	**	0.56	0.80
	Ndff† (%)	52.9	48.0	30.9	19.2	47.3	45.9	**	ns	ns
Whole Plant	N (%)	1.92	0.88	1.50	0.85	2.14	0.82	*	0.27	0.38
	Mass (g)	3.39	7.70	2.91	9.56	3.02	10.0	**	0.71	1.00
	Ndff† (%)	57.9	53.9	40.2	28.5	61.1	54.0	**	2.34	3.31
	Shoot/Root	1.35	0.97	0.91	0.48	1.87	0.85	**	0.28	0.40

*, **, ns significant at $p < 0.05$, 0.01, and nonsignificant, respectively

† N derived from fertilizer

DISCUSSION

S^o and N dynamics in nonplanted soil

In the HLS treatment, there was a rapid drop in S^o, a decline in pH, and strong correlation between N flush size and S^o oxidation. This suggests S^o acted as an energy source for the microbial population, and therefore, the presence of autotrophic S oxidizers. The relative proportions of autotrophic and heterotrophic S oxidizers were not determined here. However, previous studies have demonstrated that the forest floor near the S^o block had: high counts ($>10^5$ g^{-1}) of autotrophic *Thiobacillus thiooxidans* and *T. thioparus*, a low population of heterotrophic S oxidizers, and a higher rate of S^o oxidation for autotrophs (Gal, 1986; Germida et al., 1985; Maynard et al., 1986). Also, liming of forest soil was found to favor bacteria over fungi (Jones and Richards, 1977). *Thiobacillus sp.* were probably prime components of the microbial biomass in the HLS treatment, and to a lesser extent, in the MLS treatment.

The stimulation in N flush may not be entirely due to autotrophs. Lawrence and Germida (1988) found a positive relationship between microbial biomass C and S^o oxidation in an agricultural soil where S oxidation was dominated by heterotrophs. In our experiment, the heterotrophic population could have increased because improved access to available C was probably obtained when the soil was sieved and mixed.

All three treatments showed net immobilization of added NH_4^+, but the size and rate of immobilization were greatest in the HLS treatment. The low amount of mineral N initially present and the positive response of the flush to fertilizer N implies the availability of N could have limited the size of the soil microbial biomass in this treatment. With an ample supply of S^o, N would have been needed for autotrophic S

oxidizer growth. However the temperature and moisture conditions used favored S^o oxidiation and N immobilization, so without an unfertilized control it is not clear whether or not N was limiting. The faster immobilization of N in the HLS treatment was demonstrated by a relatively rapid depletion of the size and % ^{15}N excess of NH_4^+-N, greater buildup of labelled N in the residual fraction, and higher initial ^{15}N enrichment of the SON. Our work agrees with other studies which showed that liming of acid forest soils resulted in a net increase in immobilization, or a net decrease in mineralization, of N (Jones and Richards, 1977; Nommik, 1978; Lohm et al., 1984).

The lack of positive response of flush in the control and MLS treatments implies that the microbial biomass required lesser amounts of N than in the HLS treatment. The MLS treatment tended to have more labelled and unlabelled N in soluble forms relative to the control, suggesting a lower immobilization rate of fertilizer N. The lower rate of N immobilization in the more acid control and MLS treatments is consistent with the results of Lohm et al. (1984).

Conversion of N flush figures to biomass C allowed comparison with previous work on the upper mineral layers of acid forest soils. A k_N value represents the portion of the biomass N released by chloroform fumigation. Applying a k_N of 0.54 (Brookes et al., 1985), and a biomass C/N ratio of 6.6 (Vance et al., 1987), biomass C ranged from 50 to 510 mg C kg^{-1} soil. As determined by $CHCl_3$ fumigation, reported values for biomass C range from 20 (David et al., 1982) to 580 (Vance et al., 1987) mg kg^{-1}. Visser (1984) examined the site used in our study and found 1600 to 9300 mg of biomass C kg^{-1} in the F-H layers and 280 to 330 mg of biomass C kg^{-1} in the mineral soil. Thus the size of the microbial biomass found in our study is in general agreement with other published results.

Nitrate levels were low in this experiment, and greatest in the HLS treatment. These results are typical of other work. In the control and the MLS treatments little NO_3^- accumulated. This agrees with research summarized by Robertson (1982), which indicated NO_3^- production during incubation of northern Boreal forest soils is often less than 5 mg kg^{-1}. The latent accumulation of NO_3^- in the HLS treatment is not unusual, as pH increases caused by liming are known to promote nitrification after a delay period (Keeney, 1980; Robertson, 1982). The HLS treatment then, has the greatest potential for nitrification and subsequent losses of N.

Soluble organic N, which is probably composed of root exudates, extracellular microbial enzymes, and microbial biomass N (Van Cleve and White, 1980), is considered a source of readily available N (Weber and Van Cleve, 1984). Our study provided some support for this concept: SON levels fluctuated, and after 2 weeks, had a higher % ^{15}N excess than the residual fraction. The SON and N flush were inversely related in the control and the MLS treatments. A possible reason is that N was released to the SON pool when organisms died, and during phases of growth, SON was used as a substrate. On the other hand, in a field study, Van Cleve and White (1980) suggested high SON and biomass levels corresponded because more microbial tissue would be extracted. In support of this, $SO^{15}N$ and flush ^{15}N excesses were positively correlated. The SON fraction appears to be more readily available to the microbial biomass than residual organic N, but a simple relationship between biomass N and SON was not found in this experiment.

Adjusted unaccounted for labelled N values ranged from 0 to 6%. The adjusted values are more likely representative of actual losses than unadjusted for two reasons: 1) literature summarized by Pluth and Nommik (1981) indicates acid forest soils have low denitrification capacities; and 2) losses did not increase after the first sampling time,

even though conditions were similar throughout the experiment. The pots were sealed at the bottom; thus the only mechanism of N loss was through gaseous forms. Volatilization of NH_3 is insignificant when an NH_4^+ source is added to acid forest soils (Keeney, 1980), so losses are presumed to have been due to denitrification. As the moisture contents did not exceed field capacity, it is not suprising that the amount of denitrification was small.

Unrecovered ^{15}N was slightly higher in the HLS treatment. The greater denitrification potential of this soil was indicated by a higher: pH, C content, and accumulation of NO_3^-.

Plant growth and the influence of plants

The presence of plants did not mask the effects of the S^o and lime treatments on N dynamics. As with the nonplanted soils, the HLS treatment had the greatest rate of S^o oxidation, the highest recoveries in the flush and residual fractions, and slightly greater losses of labelled N. This indicates immobilization reduced the availability of fertilizer N to plants grown in the HLS treatment. Smaller shoot to root ratios and lower shoot yields provide further evidence that plants grown in the HLS treatment were under greater N stress. At 14 weeks the plant N content was not significantly different among the three soils. Thus a relative improvement in plant available N occurred in the HLS treatment. This could be explained by the decline in flush, which may have released plant available N, and/or increased mineralization of soil organic matter. On a mass basis, the MLS treatment had the most labelled N recovered in plants, the least in residual form, and, at 8 weeks, a tendency towards more unlabelled N in soluble forms. This implies immobilization of N was lower than the other two soils. The relative rate of N immobilization among the three soils was the same in nonplanted and planted soil.

Generally, values from planted soil were significantly lower than those from nonplanted soil (Table 2.4). Plant uptake was the cause of lower concentrations of N forms in the soil. The slightly lower pH could have been caused by plant release of H^+ to compensate for NH_4^+ uptake (Van Cleve and Moore, 1978). Elemental S content was slightly higher in planted soil. Lettl (1981, 1984) has suggested the rhizosphere is an unfavorable environment for autotrophic S oxidizers. Reduced water or nutrient availability may have accounted for the inhibition of S^o oxidation in planted pots. When extra N was added to 2 pots per treatment at 12 weeks, S^o content at 14 weeks tended to be lower compared to soil without extra fertilizer (unpublished data). This suggests that N increased the oxidation of S^o. The influence of plants on loss of labelled N could not be determined due to variability in recoveries. Other workers have found that roots promote denitrification (Klemedtsson et al., 1987), but when NO_3^- levels are low the effect of roots is reversed (Smith and Tiedje, 1979).

CONCLUSION

Of the three treatments investigated, the one with the highest amounts of S^o and lime immobilized the largest proportion of labelled fertilizer N. In addition, the lowest plant uptake of added N occurred in this soil. This study suggests that in the short term greater quantities of fertilizer N may be required when reclaiming soils with extremely high amounts of S^o and lime. If the conditions change, for example the supply of S^c is exhausted, some of the added N will be remineralized and the amounts of fertilizer N applied can be reduced.

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3. FIELD EXPERIMENT

INTRODUCTION

The previous chapter has demonstrated that increased N immobilization is partially responsible for the mineral N shortfall in limed S^o-affected soil under controlled conditions. In this chapter the reasons for the rapid disappearance of mineral N were further investigated in field experiments. Our first hypothesis, derived from chapter 2, was that more added mineral N would be immobilized in soil with higher levels of S^o and lime. Secondly, disturbance to forest ecosystems is known to cause losses of N, usually in the form of NO_3^- (Tamm, 1982; Vitousek et al., 1979). This lead to the formulation of our second hypothesis: that retention of fertilizer N in plants and the upper soil layers would be greater for NH_4^+ than for NO_3^- fertilizer. These hypotheses were tested in a field experiment by applying $(^{15}NH_4)_2SO_4$ and $K^{15}NO_3$ to forest soil with four different levels of S^o and lime.

MATERIALS AND METHODS

Study area description

The study site was located at a gas plant (52°13'N, 115°10'W), 50 km southwest of Rocky Mountain House, Alberta. Soils and geology of the area have been described by: Addison et al. (1984), Alberta Energy and Natural Resources (1984), Maynard et al. (1986), and Peters and Bowser (1960). The area borders the Alberta foothills and is part of a large morainal plain interrupted by glacial meltwater channels. Locally, the site is on a relatively flat (slope < 2%) morainal plateau at an elevation of 1204 m. The soils used were Brunisolic Gray Luvisols (Canada Soil Survey Committee, 1978) that developed on a 20 cm aeolian veneer of silt loam texture, underlain by a clay loam glacial till. The C:N ratio of the organic horizons averaged 42, and the average depths of the LF and H horizons were 2.5 and 3.5 cm, respectively. The soils used in this study were similar, except that varying additions of S^0 and lime occurred during the 5 years prior to this experiment.

Vegetation of the site belonged to the Lower Foothills section of the Boreal forest (Rowe, 1972), or the Boreal Foothills ecoregion (Strong and Leggat, 1981). Local vegetation was examined by Addison et al. (1984), Kennedy et al. (1985), and Maynard et al. (1983). Fifty year old, 25 m high *Pinus contorta* Loudon var. *latifolia* Engelm. trees dominate the canopy and *Populus tremuloides* Michx., *Picea glauca* (Moench)Voss, and *Picea mariana* (Mill) BSP are also present. *Vaccinium myrtilloides* Michx., *Linnaea borealis* L. var. *americana* (Forbes) Rehd., *Calamagrostis canadensis* (Michx.) Beauv., and *Cornus canadensis* L. are common understory components. Vascular understory plant and moss covers were 10 and 16% respectively. Near the S block total vegetative cover and diversity were significantly reduced.

The climate of the region is continental Boreal-Cordilleran; mean temperatures are -10°C in winter and 11.5°C in summer (Strong and Leggat, 1981). Thirty to 75 frost free days occur each year (Alberta Energy and Natural Resources, 1984). Total mean annual precipitation is 606 mm (439 mm as rain), 70% of which falls from May to September (Atmospheric Environment Service, 1982). The research area received 219 mm of precipitation during the study period of June through August, 1985. This was 70% of the norm.

Experimental design

Four treatments were used (Table 3.1). The control represented a soil relatively unimpinged by S^o and without added lime. The other 3 treatments were near the S^o block and were designated as: air applied lime (ALS), high lime and high S^o (HLS), and no lime (NLS). These treatments had been exposed to substantial S^o dusting for approximately 5 years. The ALS and HLS treatments had received several additions of coarsely (71% > 0.24 mm) ground limestone ($CaCO_3$) applied by aircraft from 1980 to 1984. In addition, the HLS received an extra hand broadcast application of finely ground $CaCO_3$ (19,830 kg ha^{-1}; 8% > 0.24 mm) and finely divided S^o (2,240 kg ha^{-1}) in the fall of 1980 (Gal, 1986). The NLS treatment received no lime, yet the pH was similar to the ALS treatment.

The experiment was implemented in early June 1985. Circular open ended metal cylinders (38.7 cm in diameter, 22.9 cm deep) were inserted and served to delineate the area of fertilizer application. A 250 mL solution containing 90 kg ha^{-1} of labelled N (5% atom excess) was applied. The fertilizer was spread evenly via pipette to within 4 cm of the cylinder edge. Similarly, 250 mL of distilled H_2O were applied to move the

Table 3.1. Treatments for Field Experiment

Treatment	Distance to S^o Block	Additions		S^o †	pH † of LFH	Fertilizer Applied
		S^o	Lime			
	m	t ha^{-1}	t ha^{-1}	t ha^{-1}		
1. Control Control	1500	None	None	0.05	3.9	$(NH_4)_2SO_4$ KNO_3
2. Air Applied Lime (ALS) Air Applied Lime	300	Windblown	22.1‡	2.3	3.0	$(NH_4)_2SO_4$ KNO_3
3. High Lime, High S^o (HLS)	250	Windblown + 2.2 t ha^{-1} §	22.1‡ + 19.8 §	2.7	6.1	$(NH_4)_2SO_4$
4. No Lime (NLS)	250	Windblown	None	2.7	2.9	$(NH_4)_2SO_4$

† 80 days after fertilizer application

‡ coarse limestone applied by air.

§ finely divided, applied by hand.

fertilizer into the forest floor. For each treatment $(^{15}NH_4)_2SO_4$ was applied to 3 cylinders (replicates). Thus the fate of NH_4^+ fertilizer was compared at 4 different levels of S^o and lime. For the control and ALS treatments, a further 3 cylinders received $K^{15}NO_3$. This allowed a comparison of NH_4^+ and NO_3^- fertilizers. The NH_4^+ and NO_3^- experiment had a randomized complete block design with 5 m between each of the 3 replicates. The layouts for the NH_4^+ versus NO_3^- experiment, and the influence of S^o and lime on NH_4^+ experiment, are summarized in Table 3.1. A grass seed mix of acid tolerant plants was broadcast in all cylinders at 42 kg ha^{-1}. The composition by weight was: 50% *Festuca rubra rubra*, 30% *Phleum pratense* L., 10% *Poa compressa* L., and 10% *Lolium perenne* L..

Field sampling

The soil within each cylinder was destructively sampled 80 days after fertilizer application. Natural and seeded plants were not separated and were clipped at ground level. The following 3 layers were excavated separately: LF (organic), H (organic), and 0 to 10 cm of mineral soil. Large samples were mixed, weighed, and a representative subsample taken. The 10 to 25 cm layer was sampled by taking 4 cores 3.8 cm in diameter. For the 25 to 55 cm layer, two cores 1.9 cm in diameter were removed. Samples were transported in a cooler and stored at -20°C. Average bulk density values for the LF, H, 0 to 10, 10 to 25, and 25 to 55 cm layers were: 0.1, 0.2, 0.8, 1.2, and 1.8 Mg m^{-3} respectively. A separate bulk density for each sample in the top three layers was used when converting to kg ha^{-1}. For the lower two layers the average bulk density was applied.

Analytical procedures

Soil was extracted with 2M KCl (ratio of 1g soil:15mL solution for organic material, 1g soil:3mL solution for mineral soil; shaken for 1 hr). Extracts were suction filtered through Whatman no. 42 filter paper and frozen at -15°C. Non-extracted soil was air dried and ground. Plant material was dried at 70°C, weighed, and ground. Elemental S was determined by the procedure of Hart (1961), with one exception. The organic layers of the control site were low in S^o, consequently the more sensitive method of Maynard and Addison (1985) was employed.

Steam distillation (Bremner, 1965) was used for separate determination of NH_4^+ and NO_3^-. These distillates were dried and saved for ^{15}N analysis. Total N and ^{15}N abundance were measured by combustion of air dried finely ground soil and plant samples in a Sira 1500 Automatic N analyzer coupled with a V. G. Isogas continuous flow mass spectrometer. Soil pH was measured in 0.01M $CaCl_2$ (ratio of 1g soil:6mL solution for organic material, 1g soil:2mL solution for mineral soil).

Statistical analysis

ANOVA was performed using the UANOVA version of SPSSx at the University of Alberta (Taerum, 1985). The NH_4^+ versus NO_3^- experiment was analyzed as a randomized complete block design. The NH_4^+ in four different levels of S^o and lime experiment was a two way design (treatment x depth). Since depth measurements were not independent of one another, one of the assumptions of the ANOVA was violated. This problem was overcome by the use of Greenhouse-Geisser adjusted F ratios (Greenhouse and Geisser, 1959).

RESULTS

Fate of NH_4^+ versus NO_3^- fertilizer

The fate of N was compared between $K^{15}NO_3$ and $(^{15}NH_4)_2SO_4$ fertilized soil for the control and ALS treatments only. Eighty days after fertilizer addition the NH_4^+ fertilized soil had an average of 64 kg ha^{-1} of nonlabelled mineral N (NH_4^+-N plus NO_3^--N) compared to only 28 kg ha^{-1} in the NO_3^- fertilized soil (Table 3.2). Mineral N in the NH_4^+ fertilized treatments was mostly found in the H horizon and upper 25 cm of mineral soil. In contrast, both mineral N forms in the KNO_3 fertilized soil were either relatively evenly distributed with depth, or slightly greater in the lowest soil layer. For both fertilizer types, on a concentration basis, most of the mineral N was found in the organic horizons (control, 185 to 357 mg kg^{-1}; ALS 35 to 81 mg kg^{-1}) and dropped off rapidly with depth (from 1 to 51 mg kg^{-1} in the mineral horizons) (Table A.2). Although the main treatment effect was not significant, generally the interaction effects involving treatment were (Table 3.2). The treatment by fertilizer interaction shows that the difference in the amount of NH_4^+ between the two fertilizer sources was greater in the ALS than the control treatment. Most of the mineral N was found deeper in the ALS than the control treatment (significant treatment by depth interaction).

Patterns of depth distribution (Figure 3.1a to 3.1d) and statistics (Table 3.3) for recovery of ^{15}N fertilizer were similar to those for kg ha^{-1} of nonlabelled mineral N. The difference in recoveries between NH_4^+ and NO_3^- fertilizers were strongest in the H and 0 - 10 cm layers, where the recoveries were 3 to 4 times larger in the NH_4^+ fertilized soil (Figure 3.1a to 3.1d). In the lowest soil layer sampled (25 to 55 cm) the highest recoveries were found in the NO_3^- fertilized soil. For both NH_4^+ and NO_3^-

Table 3.2. Nonlabelled mineral N and pH 80 days after fertilizer application.

Layer‡	Mineral N † NH_4^+-N	Mineral N † NO_3^--N	pH	Mineral N † NH_4^+-N	Mineral N † NO_3^--N	pH
	kg ha^{-1}			kg ha^{-1}		
	NO_3^- Fertilized					
	Control Treatment			ALS Treatment		
LF	1.6 (0.9)	0.5 (0.6)	4.1	0.7 (0.1)	0.1 (0.0)	3.2
H	6.7 (2.7)	1.0 (0.5)	3.7	2.1 (1.0)	0.4 (0.2)	2.7
0 - 10	4.7 (2.7)	1.2 (0.5)	4.6	5.3 (1.4)	2.9 (0.5)	4.0
10 - 25	2.8 (0.8)	1.2 (0.6)	4.7	4.4 (2.2)	3.3 (1.6)	4.3
25 - 55	4.8 (0.5)	3.0 (2.0)	4.5	3.5 (1.4)	4.8 (3.4)	4.6
TOTAL	20.6 (5.8)	7.1 (2.6)		16.0 (5.0)	11.5 (3.5)	
	NH_4^+ Fertilized					
	Control Treatment			ALS Treatment		
LF	3.7 (1.2)	0.7 (0.1)	4.0	0.7 (0.2)	0.1 (0.0)	3.1
H	14.0 (1.6)	5.4 (0.6)	3.9	3.1 (1.1)	0.7 (0.5)	2.6
0 - 10	18.1 (4.0)	6.8 (2.2)	4.5	26.6 (2.7)	10.7 (5.5)	4.1
10 - 25	2.8 (1.3)	1.2 (0.5)	4.7	11.3 (1.3)	6.3 (0.8)	4.5
25 - 55	4.8 (2.2)	1.3 (0.9)	4.6	5.9 (1.9)	2.8 (1.0)	4.7
TOTAL	43.5 (6.1)	15.4 (1.5)		47.5 (3.1)	20.6 (5.1)	
	NH_4^+ Fertilized					
	HLS Treatment			NLS Treatment		
LF	0.7 (0.5)	0.1 (0.0)	6.1	0.2 (0.1)	0.1 (0.0)	3.3
H	8.2 (3.8)	0.7 (0.[illegible])	6.1	2.9 (0.8)	1.3 (0.3)	2.6
0 - 10	13.9 (4.1)	5.0 (1.9)	4.6	19.6 (5.8)	14.3 (6.2)	3.9
10 - 25	4.6 (1.5)	1.6 (0.5)	4.7	18.0 (6.6)	8.0 (4.6)	4.0
25 - 55	5.3 (2.6)	1.5 (1.5)	4.7	9.3 (3.7)	4.9 (1.0)	4.6
TOTAL	32.8 (2.6)	8.8 (0.6)		50.1 (8.4)	28.6 (8.9)	

Statistical Analysis

NH_4^+ vs NO_3^- fertilizer	NH_4^+-N	NO_3^--N
Treatment	ns	ns
Fertilizer	**	**
Depth	**	**
Treat x Fertilizer	**	ns
Treat x Depth	**	*
Fertilizer x Depth	**	*
Treat x Fert x Depth	**	ns
Block effects	ns	ns

NH_4^+ fertilizer at 4 levels of S and lime	NH_4^+-N	NO_3^--N
Treatment	*	**
Depth	**	**
Treatment x Depth	**	*
Treatment x Rep	ns	ns
LSD Treat ($p<0.05$)	12.1	11.1

*, **, ns significant at $p < 0.01$, 0.05, and not significant, respectively.

† standard deviations in parentheses.

‡ soil depth in cm.

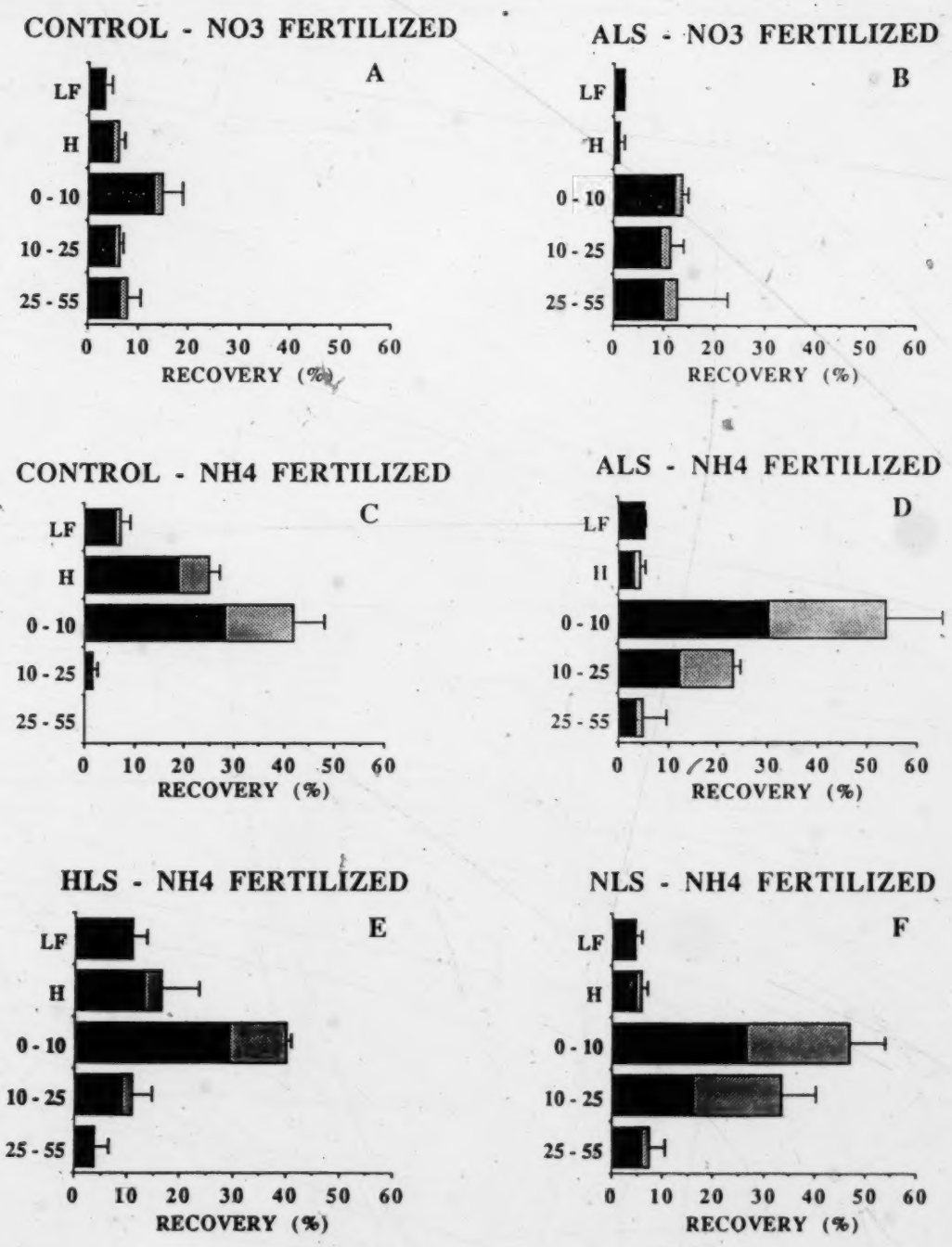

Figure 3.1. Distribution of labelled N recovered in soil.

■ Labelled immobilized N ■ Labelled mineral N

Thin bars are standard deviations. Soil depths are in cm.

Statistics are in Table 3.3.

Table 3.3. Percent recovery of labelled N in soil and plants.

N form	Recovery of ^{15}N (%) †	
	NO_3^- Fertilized	
	Control	ALS
NH_4^+	3.5 (0.8)	2.1 (0.6)
NO_3^-	1.8 (0.9)	3.6 (2.9)
Immobilized N ‡	33.0 (1.2)	35.1 (8.5)
Plants	2.0 (0.6)	0.0 (0.0)
Total Recovery	40.2 (2.0)	40.8 (11.8)
	NH_4^+ Fertilized	
	Control	ALS
NH_4^+	16.0 (0.7)	27.1 (4.5)
NO_3^-	5.1 (0.8)	9.7 (2.9)
Immobilized N	55.1 (5.3)	55.0 (5.4)
Plants	3.7 (2.9)	0.0 (0.0)
Total Recovery	79.8 (6.4)	91.8 (12.4)
	NH_4^+ Fertilized	
	HLS	NLS
NH_4^+	12.9 (2.0)	26.8 (8.5)
NO_3^-	3.2 (0.9)	13.1 (5.3)
Immobilized N	65.7 (4.7)	58.4 (3.5)
Plants	3.2 (2.4)	0.0 (0.0)
Total Recovery	85.0 (4.3)	98.4 (15.5)

Statistics for recovery of fertilizer N				
	NH_4^+	NO_3^-	Immobilized	Total
	NH_4^+ vs NO_3^- fertilizer			
Treatment	*	ns	ns	ns
Fertilizer	**	**	**	**
Depth	**	**	**	**
Treatment x Fertilizer	**	**	**	ns
Treatment x Depth	**	*	**	**
Fertilizer x Depth	**	*	**	**
Treat. x Fert. x Depth	**	ns	ns	*
Block Effects	ns	ns	ns	ns
NH_4^+ fertilizer at 4 different levels of S^o and lime				
Treatment	**	**	ns	ns
Depth	**	**	**	**
Treatment x Depth	**	**	**	**
Treatment x Rep	ns	ns	ns	ns
LSD Treat. ($p < 0.05$)	9.0	6.1	ns	ns

*, **, ns significant at $p \leq 0.05$, 0.01, and nonsignificant, respectively

† Standard deviations in parentheses

‡ Difference between total soil N and mineral N recoveries

sources of N, most ^{15}N was recovered in the 0 to 10 cm layer. There was a marked difference in total recovery of labelled N between NH_4^+ (86%) and NO_3^- (40%) fertilizers (Table 3.3). In addition, immobilization of ^{15}N was significantly lower in NO_3^- fertilized soil (34 versus 55%).

Plants grew on the control but not the ALS treatment. Plant yields (5 to 8 kg ha^{-1}) and recovery of ^{15}N (< 4%, Table 3.3) were low and not significantly different between NH_4^+ and NO_3^- fertilized plants. Percent N content was higher ($p \leq 0.01$) in NH_4^+ (2.1%) versus NO_3^- (1.7%) fertilized plants.

Influence of S^o and lime

The fate of N from $(NH_4)_2SO_4$ was compared among the 4 different levels of S^o and lime. Generally, there was a tendency for the amounts of nonlabelled NH_4^+ and NO_3^-, to decrease in the order: NLS = ALS, control, and HLS (Table 3.2). The order was the same when going from lowest to highest for pH (Table 3.2). The more acid ALS and NLS treatments had the greatest amounts of NH_4^+ and NO_3^- below 10 cm (Tables 3.2 and A.2).

Nitrate comprised 36, 30, 26, and 21% of the mineral N in the NLS, ALS, control, and HLS treatments respectively. Nitrate was found in the 0 - 10 cm layer of all NH_4^+ fertilized treatments, even though the pH was approximately 4 (Table 3.2). The pH of the organic layers was less than in the mineral soil, except for the HLS treatment (Table 3.2). Thus if NO_3^- was produced in the LFH and leached to the mineral soil, nitrification took place at an even lower pH than in the 0 - 10 cm layer. The most acid organic layer (pH 3.5) where appreciable NO_3^- occurred was the H horizon of the control.

Total recovery of ^{15}N ranged from 80 to 98%, although differences among the 4 treatments were not significant ($p = 0.2$) (Table 3.3). However, the distribution of recovered fertilizer N within the profile (Figure 3.1c to 3.1f), varied among the treatments. The control and HLS treatments retained more ^{15}N in the organic layers than the ALS and NLS treatments. Immobilized ^{15}N was highest (66%) in the HLS treatment (Table 3.3). When immobilization was given as a percent of labelled N recovered in the soil, rather than a percent of the amount added, it increased, again in the order of NLS = ALS < control < HLS (LSD $p \leq 0.05$). Of the ^{15}N remaining in the soil, 60 to 80% was immobilized. In all treatments the largest recoveries were found in the 0 - 10 cm layer of mineral soil (Figure 3.1c to 3.1f). Below this depth, the highest levels of recovered ^{15}N (28 to 41%) were in the more acid ALS and NLS treatments. In contrast, in the control, less than 2% of added labelled N was recovered below 10 cm.

For the NH_4^+ fertilized treatments, plants grew only on the control and HLS treatments. Plant yields (6 to 8 kg ha^{-1}) and ^{15}N recovery (3 to 4%, Table 3.3) were low. There was no significant difference between the two treatments for yield, ^{15}N content, or N concentration.

DISCUSSION

Fate of NH_4^+ versus NO_3^- fertilizer

Eighty days after fertilizer application, total recovered labelled N to a depth of 55 cm was 86% in NH_4^+ fertilized soil versus only 40% with NO_3^- fertilizer. This 46% difference relates well with similar work done in a comparable time frame; Nommik and Popovic (1971) found a 25% difference, and Overrein (1971a) found 67% more leaching to a depth of 40 cm with NO_3^- fertilizer. The difference in vertical distribution between NH_4^+ fertilizer and the more mobile NO_3^- fertilizer is well known (Nommik and Popovic, 1971, Melin and Nommik, 1988, Overrein, 1970, 1972). Our results agreed with these studies in that: 1) NH_4^+ fertilizer was partially retained in the organic layers, and showed limited movement below the upper mineral soil, and 2) NO_3^- fertilizer was leached into the lower layers of mineral soil and little was immobilized. The poor retention of NO_3^- fertilizer has negative long term implications for plant growth, because less mineral N was retained in the upper soil. This was reflected to some extent in the plant data, even though the experimental period was short. The NH_4^+ fertilized plants had a slightly higher concentration of N, probably because NO_3^- was lost from the rooting zone before the seedlings could absorb it. The poor growth and small plot size make these conclusions tentative. Nevertheless, in our study, NH_4^+ fertilizer was a more effective N source than NO_3^- because more was retained in the system, both in total and in the upper soil layers, and greater amounts of mineral N were available for plant growth.

The unrecovered NO_3^- fertilizer could have been lost by leaching and/or gaseous evolution. Moisture data showed that the wet conditions which promote both these mechanisms of N removal were prevalent during the experiment. The total rainfall of

219 mm suggested anaerobic conditions were possible at certain times, and generally soil moisture was at or above field capacity (33 kPa) when sampled (Fig. A.1 and Table A.2). Leaching below the zone of sampling is suggested by the 8 to 12% recovery of labelled N in the lowest soil layer sampled (25 to 55 cm). At a another nearby S^o-laden site, SO_4^{-2} was leached below this depth (Maynard, pers. comm.). Nitrate is more mobile than SO_4^{-2}, implying NO_3^- moved below 55 cm. Gaseous N losses from NH_4^+ and NO_3^- fertilized acid forest soils are considered to be low (Keeney, 1980). Van Pragg and Weissen (1984) however, found 10% of applied NO_3^- was lost in gaseous forms after one month. In addition, the amount of denitrification can become equivalent to leaching losses when promoted by the elevated NO_3^- levels which often occur following disturbance (Robertson et al., 1987). In the lowest layer sampled most of the pore space was filled with water; this would increase the potential for denitrification in this layer. Although the mechanism of N loss remains uncertain, evidence suggests leaching played a larger role.

Influence of S^o and lime

The greatest amount of ^{15}N was immobilized in the treatment with the highest level of S^o and lime (HLS). While both the HLS and ALS treatments were limed, only the finely divided lime applied to the HLS treatment was effective in raising the pH of the LFH. Liming usually has a positive effect on N immobilization (Nommik, 1978; Popovic, 1984; Tamm et al., 1977). The higher immobilization in the HLS was confirmed in a growth chamber experiment (see chapter 2). Although more fertilizer N was retained in the upper soil layers of the HLS treatment, most of the N was immobilized. The availability of N to seedlings was probably further reduced in this treatment because roots preferred the less acid organic layer, even though more N was available in the mineral soil. Thus in the short term, soils with high levels of S^o and

lime will require more mineral N, because much of the mineral N is converted to forms which are unavailable to plants. In the long term, this N may be slowly released, especially if the supply of S^0 becomes limited.

In the control, the lack of labelled N recovered below 10 cm suggests leaching was less important than denitrification as a potential mechanism of N removal. This treatment retained more fertilizer N in the organic layers, where denitrification can be more prevalent than in the mineral soil (Federer and Klemedtsson, 1988).

The NLS and ALS treatments had the lowest pH and amount of immobilized N, and the highest levels of mineral N and ^{15}N recovered below 10 cm (Tables 3.2, 3.3, and Figure 3.1). Acid additions to coniferous forest soils sometimes reduce the size, or shift the composition of, the microbial community, resulting in impaired nutrient cycling (Baath et al., 1980, 1984; Berg, 1986; Bewley and Parkinson, 1983; Lettl, 1984; Maynard et al., 1986). Thus the lower amount of N immobilization in the more acid NLS and ALS treatments is not surprising, and agrees with Tamm et al. (1977). Leaching of N was enhanced in these treatments because: 1) the high concentration of H^+ can prevent NH_4^+ fixation (Overrein, 1969), and 2) more N was present as NO_3^-. The accumulation of NO_3^- at low pH is not common, but has been previously observed (Klein et al., 1983; Overrein, 1971b), especially when vegetation uptake is minimal (Tamm, 1982). Therefore nitrification is a potentially important process which could increase N loss in these acidified soils.

CONCLUSIONS AND IMPLICATIONS

Soil recovery of fertilizer NH_4^+ was greater than NO_3^-. Therefore an NH_4^+ based fertilizer should be more effective in reclaiming the affected area because more N was recovered in the potential rooting zone. After 80 days plant yield and N uptake were too low to warrant conclusions about treatment effects.

The amount of N immobilization was large and was greatest in the soil with the most S^o and lime. Consequently, in the short term, higher amounts and/or more frequent additions of fertilizer N may be justified when reclaiming soils with high amounts of S^o and lime. In the long term, this N may be slowly released.

Nitrification occurred in these acid forest soils following NH_4^+ addition. This suggests that potential losses of N could be reduced by using smaller more frequent N additions, rather than a single large dose.

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4. SYNTHESIS

This study examined the fate of fertilizer N in S^o-laden forest soils. Experiments were performed in a growth chamber and in the field. The salient points of this research are as follows:

1. Mineralization-Immobilization

Conclusion: Immobilization of N was greatest in the soil with the most S^o and lime.

High amounts of NH_4^+ fertilizer were immobilized. In the soil with the most S^o and lime, immobilization of N was probably enhanced by autotrophic S oxidizers. The latter theory could be strengthened if autotrophic and heterotrophic populations were enumerated.

Implications: Mineral N from fertilizer is less available to plants in soils with high amounts of S^o and lime. Consequently, in the short term, higher amounts and/or more frequent additions of fertilizer N may be warranted. Plants with low N requirements, preferably native ones, should be considered. In the long term, the immobilized N may be slowly released, especially if the supply of S^o is exhausted. This was demonstrated in the incubation experiment.

2. Fertilizer form

Conclusion: $(NH_4)_2SO_4$ was a more effective N carrier than KNO_3.

NO_3^- fertilizer was lost from the upper soil layers by leaching and/or conversion to gaseous forms. The amount of N loss from NO_3^- fertilizer was high, so the

mechanism of N removal for NO_3^- deserves further investigation. Other N fertilizer sources, for example urea, could be considered in future research.

Implications: An NH_4^+, rather than NO_3^- based fertilizer should be used to reclaim the affected area.

3. Nitrification

Conclusion: Use of NH_4^+ fertilizer in the acid forest soils studied does not guarantee protection against nitrification and subsequent N losses.

Nitrification occurred in these acid forest soils following NH_4^+ addition. In the field study, the treatment with finely divided lime and S^o immobilized more N, and nitrification was reduced; this helped to retain N in the upper soil. More nitrification occurred in the other field treatments. In contrast, during incubation, only the treatment with the highest level of S^o and lime produced NO_3^-. Reasons for these differences between the incubation and field experiments remain unresolved; the factors which limit nitrification in forest soils are not straightforward.

Implications: Potential losses of N could be reduced by using smaller more frequent N additions, rather than a single large application.

5. APPENDIX

Table A.1. Nonlabelled N †, S^o †, and pH in planted soil.

N form	Time (weeks) 8	14
	Control	
S^o	45	50
pH	4.4	4.4
NH_4^+-N	0.9	0.4
NO_3^--N	0.1	0.0
Flush of N‡	27.1	21.0
Soluble Organic N	5.0	5.8
	High Lime and S^o	
S^o	1170	820
pH	5.8	5.8
NH_4^+-N	1.2	0.2
NO_3^--N	0.0	0.0
Flush of N‡	33.9	23.5
Soluble Organic N	4.1	4.2
	Medium Lime and S^o	
S^o	345	190
pH	4.6	4.6
NH_4^+-N	5.0	0.0
NO_3^--N	0.1	0.1
Flush of N‡	24.4	22.6
Soluble Organic N	4.8	4.4

Statistics for planted pots

	Source of variation			LSD§ ($p < 0.05$)	
	Treatment	Time	Trt x Time	8 wks	14 wks
S^o	**	**	*	149	211
pH	**	ns	ns	ns	ns
NH_4^+-N	ns	*	ns	ns	ns
NO_3^--N	ns	ns	ns	ns	ns
Flush of N	**	**	*	3.6	5.1
Soluble Organic N	ns	ns	ns	ns	ns

*, **, ns significant at $p < 0.05$, 0.01, and nonsignificant, respectively

† mg kg^{-1} soil

‡ uncorrected for microbial biomass N

§ LSDs are for comparisons within a time only

Table A.2. Concentration of nonlabelled mineral N 80 days after fertilizer application.

Layer	Mineral N † NH_4^+-N	Mineral N † NO_3^--N	Mineral N † NH_4^+-N	Mineral N † NO_3^--N
	mg kg^{-1}		mg kg^{-1}	
	NO_3^- Fertilized			
	Control Treatment		ALS Treatment	
LF	138.6 (82.9)	46.4 (49.2)	33.0 (4.3)	4.1 (2.2)
H	199.0 (95.1)	31.0 (16.2)	53.9 (8.3)	9.6 (1.4)
0 - 10	6.3 (3.8)	1.6 (0.5)	6.9 (1.7)	3.8 (0.3)
10 - 25	1.6 (0.5)	0.7 (0.4)	2.5 (1.2)	1.8 (0.9)
25 - 55	0.9 (0.1)	0.6 (0.4)	0.6 (0.3)	0.9 (0.6)
	NH_4^+ Fertilized			
	Control Treatment		ALS Treatment	
LF	280.7 (121.6)	53.6 (19.7)	31.7 (6.4)	3.4 (0.9)
H	255.9 (41.8)	101.2 (30.3)	67.0 (11.3)	14.0 (6.7)
0 - 10	21.0 (4.1)	7.9 (2.5)	36.1 (4.1)	14.6 (7.7)
10 - 25	1.6 (0.7)	0.7 (0.3)	6.4 (0.7)	3.6 (0.4)
25 - 55	0.9 (0.4)	0.2 (0.2)	1.1 (0.4)	0.5 (0.2)
	NH_4^+ Fertilized			
	HLS Treatment		NLS Treatment	
LF	41.9 (30.9)	4.2 (1.8)	16.8 (10.0)	7.5 (1.9)
H	169.6 (69.9)	14.1 (7.3)	43.6 (18.0)	18.4 (5.9)
0 - 10	19.8 (7.2)	6.9 (1.8)	22.5 (7.2)	16.5 (7.8)
10 - 25	2.6 (0.8)	0.9 (0.3)	10.2 (3.7)	4.5 (2.6)
25 - 55	1.0 (0.5)	0.3 (0.3)	1.7 (0.7)	0.9 (0..2)

Statistical Analysis

NH_4^+ vs NO_3^- fertilizer	NH_4^+-N	NO_3^--N
Treatment	**	**
Fertilizer	*	*
Depth	**	*
Treat x Fertilizer	ns	ns
Treat x Depth	*	*
Fertilizer x Depth	ns	*
Treat x Fert x Depth	ns	*
Block effects	*	ns

NH_4^+ fertilizer at 4 levels of S and lime	NH_4^+-N	NO_3^--N
Treatment	**	**
Depth	**	**
Treatment x Depth	**	**
Treatment x Rep	ns	ns

*, **, ns significant at $p < 0.01$, 0.05, and not significant, respectively.
† standard deviations in parentheses.

Table A.3. Field capacity and moisture content at sampling time.

Treatment	Layer	% Moisture Content	
		Field Capacity†	When Sampled
Control	LF	140	210
	H	140	190
	0 - 10	40	40
High Lime, High S^o	LF	80	60
	H	105	130
	0 - 10	50	50
Air Applied Lime and S^o	LF	110	70
	H	110	160
	0 - 10	50	50
No Lime and S^o	LF	110	60
	H	120	150
	0 - 10	40	50

† Determined at a tension of 33 kPa

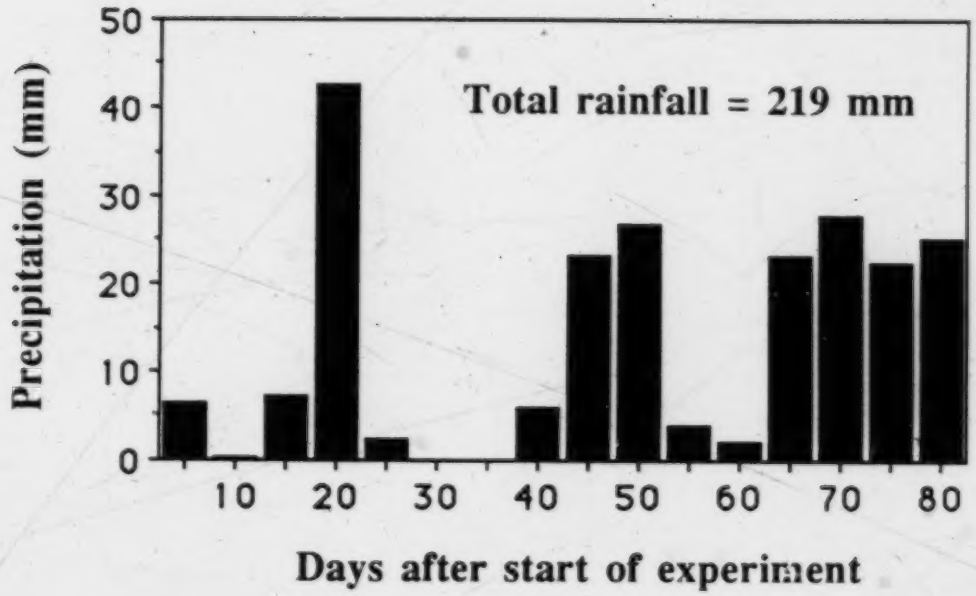

Figure A.1. Rainfall distribution near study site from June through August 1985.
Data taken from meterological station 20 km from study site.
Bars represent groupings of 5 days.